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# The Pericyte: A Forgotten Cell Type with Important Implications for Alzheimer's Disease?

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## Abstract

Pericytes are cells in the blood–brain barrier (BBB) that degenerate in Alzheimer's disease (AD), a neurodegenerative disorder characterized by early neurovascular dysfunction, elevation of amyloid β-peptide (Aβ), tau pathology and neuronal loss, leading to progressive cognitive decline and dementia. Pericytes are uniquely positioned within the neurovascular unit between endothelial cells of brain capillaries, astrocytes and neurons. Recent studies have shown that pericytes regulate key neurovascular functions including BBB formation and maintenance, vascular stability and angioarchitecture, regulation of capillary blood flow, and clearance of toxic cellular by-products necessary for normal functioning of the central nervous system (CNS). Here, we review the concept of the neurovascular unit and neurovascular functions of CNS pericytes. Next, we discuss vascular contributions to AD and review new roles of pericytes in the pathogenesis of AD such as vascular-mediated Aβ-independent neurodegeneration, regulation of Aβ clearance and contributions to tau pathology, neuronal loss and cognitive decline. We conclude that future studies should focus on molecular mechanisms and pathways underlying aberrant signal transduction between pericytes and its neighboring cells within the neurovascular unit, that is, endothelial cells, astrocytes and neurons, which could represent potential therapeutic targets to control pericyte degeneration in AD and the resulting secondary vascular and neuronal degeneration.

## INTRODUCTION

Cerebrovascular and neuronal functions are intimately entwined in the central nervous system (CNS) (75, 76, 194–196). Neurons have comparatively high metabolic rates with limited cellular reserves and require a nearly continuous supply of energy metabolites to maintain and/or replenish the ionic gradients and other metabolic functions required for rapid synaptic transmission (66). To meet these needs, neurons require continuous cerebral blood flow (CBF) for delivery of oxygen. Additionally, transport systems in brain endothelium of the blood–brain barrier (BBB) mediate the delivery of glucose and other nutrients from blood to brain, as well as the clearance of toxic metabolic by-products from the CNS back to the circulation (195, 196). To ensure adequate vascular access, the mammalian brain has evolved to become a densely vascular structure with the diffusion distance between neurons and an adjacent capillary rarely exceeding 15 μm (166). In humans, the calculated total perfused vascular length of the cerebrovascular tree is 600–700 km, with major contribution coming from dense capillary networks (194). When CBF is disrupted and/or diffusion distance for transport exchange of metabolites between neurons and circulating blood is increased, neuronal damage may occur rapidly (109).

The coordinated matching of vascular supply to neuronal demand is not simply the result of a network of passive vascular conduits. Rather, individual vascular segments adjust both CBF and transport processes at the BBB in response to systemic, neuronal and glial signals (75, 195). Appropriate vascular responses require tight and coordinated cross-talk between multiple cell types in the CNS—as reflected by the concept of the "neurovascular unit" (NVU) (177, 195, 196). At a cellular level, the NVU is composed of vascular cells [endothelial cells, vascular smooth muscle cells (vSMCs) and pericytes], glial cells (microglia and astrocytes) and neurons (Figure 1). One of these cell types—a perivascular cell known as the "pericyte"—is uniquely positioned within the NVU between vascular, neuronal and glial cells (4, 177). Pericytes are now believed to control key neurovascular functions necessary for neuronal homeostasis (14, 177). Only recently, a link between pericyte loss and/or malfunction and neurologic disease has begun to be elucidated.

Human neurodegenerative disorders, such as Alzheimer's disease (AD), have marked vascular pathophysiology, including endothelial and pericyte degeneration, instability and rupture of the vascular wall, disruption of the BBB and/or dysfunctional BBB transport systems (21, 50, 75, 196). The role of the pericyte in contributing to AD-related neurovascular dysfunction has recently been suggested (14, 15, 139, 146, 177, 196). In the present review, we first introduce the pericyte and describe evidence supporting its role in regulating the BBB, microvascular structure, blood flow

regulation and phagocytosis, and clearance of extracellular molecules from brain interstitial fluid (ISF). We then continue with the description of the vascular contributions to AD pathogenesis with an emphasis on pericyte-derived sources of injury that are both independent and dependent on the AD neurotoxin amyloid β-peptide (Aβ).

## THE PERICYTE

Pericytes are a perivascular cell population embedded on the capillary wall in a shared basement membrane with the endothelium (Figure 1) (4, 41, 177). Along the arterial–venous axis, pericytes are predominately found in capillaries, post-capillary venules and rarely terminal arterioles. According to the prevailing view, pericytes share certain characteristics with the vSMCs (4). However, the point of transition from vSMC to pericyte remains still poorly understood. Pericytes are composed of a cell body and elongated, multiple finger-like cytoplasmic processes which elongate and cover the abluminal side of brain capillaries in a manner that is very different from vSMC (41, 177). At discrete points, lacking a basement membrane, pericytes and endothelial cells form direct cell-to-cell contacts, known as "peg-and-socket" contacts. At points of contact, connexin-43 hemichannels form gap junctions permitting direct endothelial–pericyte communication (18). Fibronectin-rich adhesion plaques connect the basement membrane to the plasma membrane and underlying actin cytoskeleton in both cell types (41). Pericyte–astrocyte and pericyte–neuron contacts are much less understood. However, it appears that pericytes may play a role in guiding the association of astrocyte end-feet with the vessel wall (5).

The CNS has higher pericyte-to-endothelial cell ratios than peripheral vascular beds, that is, estimated 1:1–1:3 in the CNS vs. 1:10–100 in striated muscle, according to some early pioneering studies (148). Within the CNS, as much as 70%–80% of the capillary tube is covered with pericyte cell processes (5, 14, 15, 35, 139, 146, 178). Although coverage values are relatively consistent

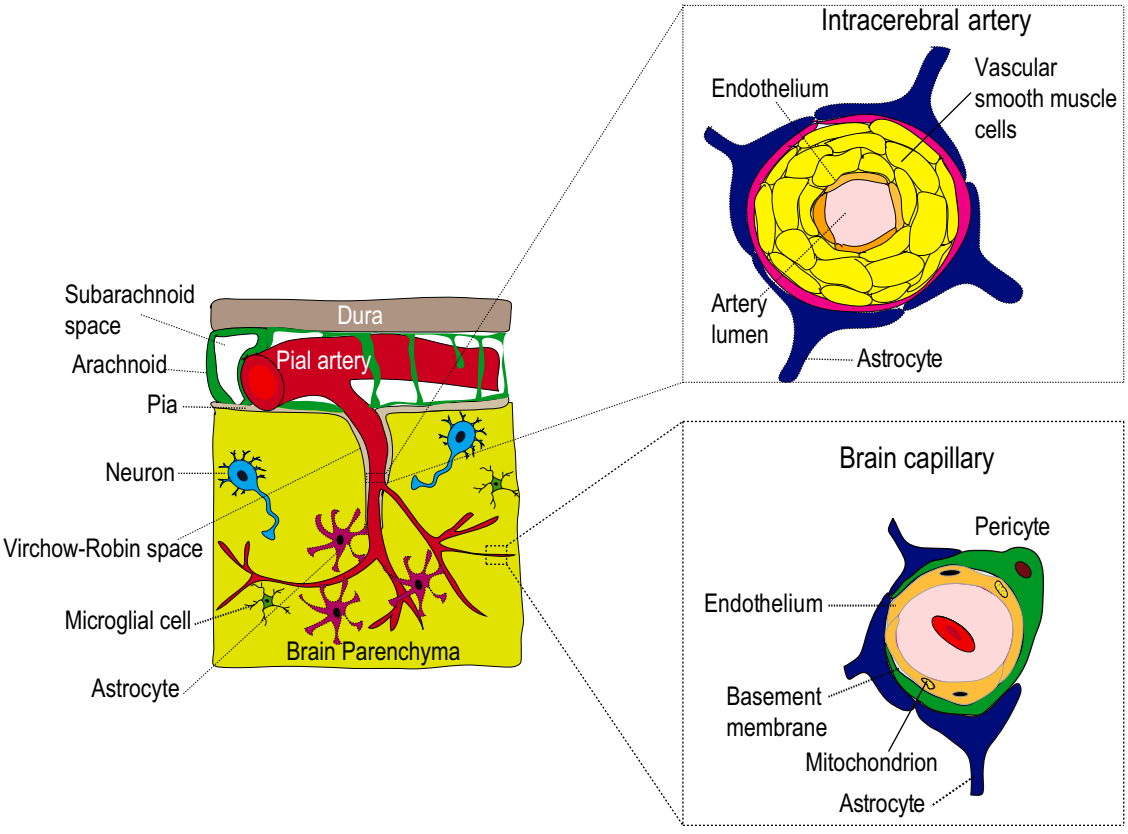


**Figure 1.** Cerebrovascular structure and the neurovascular unit (NVU). In the brain, pial arteries travel along the cerebrospinal fluid filled subarachnoid space and give rise to penetrating intracerebral arteries, which enter the brain parenchyma but are separated from neurons and glia by the perivascular Virchow-Robin spaces defined by an outer wall of pia and astrocyte-derived glial limitans membrane. These arteries branch into smaller arteries, arterioles and brain capillaries distally. At a cellular level, vascular function requires coordinated cross-talk between multiple cell types of the NVU. The NVU is composed of endothelial cells, vascular mural cells (vascular smooth muscle and pericytes), glial cells (astrocytes and microglia) and neurons. The identity of mural cells changes along the arterial–venous axis. In intracerebral arteries, vascular smooth muscle cells occupy most of the vascular wall. At the level of brain capillaries, pericytes replace smooth muscle cells and are attached to the vascular basement membrane. Pericytes extend multiple cytoplasmic processes that encircle endothelial cells. The point of transition from smooth muscle to pericyte remains poorly defined. At each level, mural cells are further surrounded by astrocyte end-feet and are in close proximity to neurons and microglia. Figure modified from Zlokovic (196).

within cortical regions across independent reports (14, 176, 178), regional heterogeneity in more permeable regions, such as the motor neuron dense regions of the spinal cord and fetal germinal matrix, has also been described (20, 178). Whether a similar relationship exists in the brain's circumventricular organs which lack a BBB has yet to be determined.

### Origin of CNS pericytes

CNS pericytes arise from several progenitor cells. During early embryonic vasculogenesis, chimerization experiments have established that neuroectoderm–mesodermal-derived progenitor cells give rise to pericytes in the forebrain and midbrain, brainstem, spinal cord and peripheral organs (49, 89, 90). Later in the embryonic period and into the early postnatal period, intraparenchymal proliferation of existing pericytes—a process termed "longitudinal expansion"—contributes to the further expansion of the brain pericyte population (1, 68, 117, 118, 155, 157). Whether longitudinal recruitment continues throughout adulthood is not well understood at present. Recent studies suggest that the cerebral microvasculature is not quiescent and undergoes continued remodeling throughout life (64). However, the turnover of pericytes within the NVU under physiological conditions has yet to be determined. In models of ischemic injury, circulating bone marrow progenitor cells have been demonstrated to contribute to brain pericyte populations (88, 92, 131). However, it has yet to be shown whether circulating progenitors may also contribute to pericyte populations in the absence of CNS injury. Additionally, the cellular source for replenishing the CNS pericyte populations during postnatal CNS development and in the adult and aging CNS is not presently known.

### Endothelial–pericyte molecular cross-talk

During vasculogenesis and/or angiogenesis, endothelial cells secrete and/or present cell-bound molecular cues that stimulate pericytes to proliferate, migrate and attach to nascent capillary tubes. This cross-talk occurs through several well-defined signal transduction cascades, including platelet-derived growth factor B (PDGF-B), transforming growth factor-β (TGF-β), NOTCH, sphingosine-1 phosphate and angiopoietin cell signaling [reviewed in (4, 54, 177)]. Each signaling cascade makes differential contributions to the process of "pericyte recruitment." In simplified form, endothelial cells secrete PDGF-BB, a homodimer of PDGF-B, which binds to and activates the platelet-derived growth factor receptor β (PDGFRβ) located on the pericyte plasma membrane, resulting in pericyte proliferation and migration (1, 54, 68, 97, 159). TGF-β and NOTCH signaling then mediate pericyte attachment via endothelial upregulation of the adhesion molecule N-cadherin (95). Bidirectional TGF-β signaling between pericytes and endothelium regulates quiescence, maturation and differentiation in both cell types (42, 150, 156, 169). Maintenance of PDGF-B/PDGFRβ and pericyte NOTCH3 signaling are required for maintenance of pericyte survival (14, 60). Recent evidence suggests that pericyte NOTCH3 may also facilitate pericyte proliferation during development (174).

### Animal models of pericyte deficiency

Genetic manipulation of endothelial–pericyte signaling pathways results in animal models of pericyte deficiency. For example, deletion and/or genetic manipulation of *Pdgfrβ* and/or *Pdgfb* result in widely utilized pericyte deficient mouse models (5, 14, 35, 159, 176). Pericyte-deficient transgenic mice have proved to be valuable tools in elucidating the functional roles of CNS pericytes *in vivo* and the effects of a chronic BBB disruption caused by pericyte degeneration on neuronal structure and function.

## NEUROVASCULAR FUNCTIONS OF CNS PERICYTES

Initially, pericytes were described as contractile cells around endothelial cells in small blood vessels by the French physiologist Rouget in 1873 (136). Over a century later, the pericyte's functional role in the CNS has been expanded through organotypic slice preparations, *in vitro* BBB models and the *in vivo* characterization of cerebrovascular and neuronal phenotypes in pericyte-deficient transgenic mice (177). Recent *in vivo* reports have established that CNS pericytes play pivotal regulatory roles in the induction and maintenance of the BBB, microvascular stability, capillary density and angiogenesis, capillary diameter, and blood flow regulation and the clearance of macromolecules from brain ISF.

### Induction and maintenance of the BBB

The BBB tightly regulates the molecular exchange between circulating peripheral plasma and the CNS. The anatomic BBB is organized at the level of a continuous endothelial layer that lines the lumen of the cerebrovascular tree. Each endothelial cell is connected to adjacent endothelial cells via tight junctional complexes and adherens junctions, which limit unregulated paracellular transport of circulating molecules. Low levels of transendothelial vesicular transport further limits non-specific transport of polar solutes and large macromolecules into the CNS (75, 195, 196). A series of tightly regulated transport systems for nutrients and energy metabolites and for brain clearance of metabolic waste products have evolved in the CNS endothelium to meet neuronal metabolic needs and maintain a microenvironment supportive of neuronal function. Large molecules, such as peptides and proteins, are in general transported slowly across the BBB via specialized transport systems (198, 200, 201) or are excluded from the brain in the absence of a specific transport system (193, 202).

Independent studies utilizing pericyte-deficient mice have demonstrated that pericytes play a pivotal role in establishing and maintaining endothelial BBB properties (5, 14, 35). Early in embryogenesis, pericytes accompany endothelial cells as they invade neural tissue from the adjacent perineural vascular plexus (35, 68). Recent work in pericyte-deficient $Pdgfr\beta^{+/-}$ mice has demonstrated that pericytes induce formation of a functional BBB prior to the appearance of perivascular astrocytes (35). Similarly, embryonic pericyte detachment leads to BBB disruption, vascular leakage and overt hemorrhage in the immediate postnatal period (95). At a molecular level, initial BBB formation is achieved through pericyte-driven downregulation of endothelial gene products which promotes permeability through transcytosis, for example, caveolin-1 (*Cav1*) and plasmalemma vesicle-associated protein (*Plvap*), and/or leukocyte infiltration, for example, intercellular adhesion molecule 1 (*Icam1*) (35, 67). Therefore, pericytes

suppress a leaky and pro-inflammatory phenotype of endothelial cells during the embryonic period.

Following birth, pericytes maintain endothelial BBB and blood–spinal cord barrier properties throughout life (5, 14, 95, 139, 178). Within the adult CNS, there is an inverse relationship between regional pericyte coverage and vascular permeability under physiologic conditions (178). In adult and aged pericyte-deficient *Pdgfrβ*$^{+/-}$ mice, a loss of brain pericytes increases non-specific paracellular endothelial transport through disrupted tight junctions (14, 178). For example, the levels of the essential tight junction transmembrane proteins occludin and claudin-5, the adaptor protein zonula occludens-1 and the adherens junction protein vascular endothelial cadherin are progressively reduced with aging in pericyte-deficient mutants. This leads to brain accumulation of exogenous vascular tracers and circulating plasma proteins with vasculotoxic and neurotoxic properties, including thrombin, fibrinogen/fibrin, plasminogen/plasmin and different non-immune and immune immunoglobulins (14). Other works in young pericyte-deficient *Pdgfb*$^{ret/ret}$ mutants has also demonstrated increased transendothelial vesicular transport. Therefore, in the adult brain, pericytes maintain BBB properties through promotion of endothelial tight junctional complexes and suppression of non-specific vesicular transport (5). Unlike the embryonic period, brain pericyte loss does not lead to upregulation of inflammatory cytokines, chemokines or enhanced immune cell infiltration in early to mid-adulthood, but only with advanced aging (14).

## Microvascular stability

Vessels that lack pericytes are dilated and tortuous with frequent rupture prone out-pouchings of the vascular wall—called "microaneurysms" (4, 48, 67, 95, 97). Evidence of rupture and/or vascular fragility is frequently evident in pericyte-deficient mutants (14, 95). In humans, brain or spinal cord pericyte reduction is associated with hemorrhage in prematurity, amyotrophic lateral sclerosis (ALS) and AD (20, 146, 179). Recent works have suggested that pericyte–endothelial cell interactions stabilize the vascular wall through multiple mechanisms. Pericytes secrete the stabilizing molecule angiopoeitin 1, which activates endothelial receptor Tie2 receptors (79). Pericytes secrete transforming growth TGF-β, which activates the TGFβR2-Alk5-Smad2/3 pathway within endothelium (3). This promotes endothelial maturation including formation of both stabilizing barrier properties and the vascular basement membrane (BM) (42, 156)—a protein-rich peri-endothelial scaffolding of extracellular matrix necessary for vascular cell attachment (195). Pericyte-endothelial signaling stimulates endothelial production of multiple BM proteins, including fibronectin, nidogen-1, perlacan and laminin (156). Pericytes also directly synthesize and secrete fibronectin contributing to BM structure and downregulate and/or inhibit the activity of destabilizing matrix metalloproteinases (MMP), including MMP-2 and MMP-9, in the latter stages of angiogenesis (144, 156). In the setting of pericyte loss, disruption of the BM contributes to vascular instability, dilatation and rupture (14).

## Angiogenesis and microvascular density

Pericytes play dynamic and at times opposing roles in angiogenesis. Early in angiogenesis, pericytes secrete angiogenic factors, that is, vascular endothelial growth factor-A (VEGF-A), which stimulates sprout formation, endothelial proliferation and/or survival (36, 118). Pericytes also express multiple MMPs, including MMP 2, 3 and 9, and urokinase plasminogen activator receptor to degrade BM proteins to facilitate endothelial migration (23, 45, 172). Some works have also suggested that pericyte migration precedes and guides endothelial tube formation, but this remains controversial (56, 117, 118, 172). Later in angiogenesis, pericytes inhibit endothelial proliferation and deposit BM proteins as described earlier. Pericytes also secrete the tissue inhibitor of metalloproteinase 3 (TIMP3) capable of inhibiting multiple MMPs, including MMP 1, 10 and 14 as well as a disintegrin and metallopeptidase domain 15 (ADAM15) (144, 156); this stabilizes the BM and concludes the angiogenic process. In pathologic states, pericyte angiogenic functions may become dysregulated with profound implications for CNS vascular structure. For example, overactivation of pericyte MMP-9 and/or loss of pericyte-derived trophic support contribute to capillary loss and ultimately hypoperfusion in adult mice (14, 15).

Continued brain angiogenesis is counterbalanced by vascular pruning throughout life (64). The net balance between these two processes regulates capillary density. Studies in pericyte-deficient mutants have revealed context-dependent effects of pericytes on regulation of capillary density. In the developing CNS, pericyte loss or detachment leads to endothelial hyperproliferation without changes in capillary branching and/or vascular density (67, 96). In the adult brain, however, variable levels of brain pericyte loss result in endothelial apoptosis, microvascular regression and reductions in capillary density (5, 14). Generation of a wide spectrum of pericyte deficient mutants through endothelial specific deletion of *Pdgfb* results in either vascular regression and/or vascular hyperproliferation depending on the magnitude of pericyte loss in the retina (48). This suggests that pericytes may have dose-dependent as well as context-dependent effects on vascular networks. However, inducible models of pericyte deficiency are required to better delineate the roles of pericytes during postnatal CNS development, growth, adulthood and aging.

## Neurovascular coupling and blood flow regulation

Blood flow is carefully matched to neuronal metabolic need in the CNS—a process called "neurovascular coupling" (7, 129). It was previously thought that blood flow modulation was predominately regulated through alterations in vSMC tone in penetrating cerebral arteries and/or arterioles in response to synaptic transmission and release of vasoactive mediators [reviewed in (75)]. This view, however, has more recently been complemented by multiple reports suggesting that pericytes may also play a role in blood flow modulation at the capillary level.

Pericytes express both contractile proteins (44, 62) and receptors for multiple vasoactive mediators including catecholamines, vasopression, angiotensin II, endothelin-1, adenosine and VIP (41, 62). Exposure of pericytes to vasoactive substance results in elevation of intracellular calcium, which initiates the contractile apparatus (69, 83, 115). In organotypic cerebellar slice and retinal preparations, pericytes were shown to alter capillary diameter by constricting or dilating in response to neurotransmitters and/or electrical stimulation (126). It was also demonstrated that

stimulation of the contractile response propagated among adjacent pericytes (126); therefore, pericytes may help coordinate responses of an entire microvascular segment. The absence and/or reductions in brain or retinal pericytes are associated with capillary dilatation *in vivo*, confirming that pericytes regulate capillary diameter *in vivo* (5, 67, 96, 97). Conversely, pericytes have been shown to assume a hypercontractile phenotype and obstruct capillary blood flow in response to pathologic injury, such as ischemia (187) and/or traumatic brain injury (46).

Despite possessing contractile properties, the relationship between pericyte-mediated capillary dilatation and/or constriction and blood flow regulation remains less definitive *in vivo*. A loss of brain pericytes in the young mouse brain results in impairment of blow flow responses to brain activation in the presence of unaltered electrophysiological neuronal responses, suggesting that pericytes fulfill an important regulatory role in functional hyperemia (14). However, an independent study confirmed pericyte-mediated constriction of cortical capillaries, but suggested that this did not alter functional hyperemic responses *in vivo* (52). This raises questions as to the significance of capillary diameter changes and its relevance to modulating blood flow responses to brain activation. More recently, carefully designed *in vivo* multiphoton experiments have suggested that pericytes dilate before arterioles in response to neuronal stimulation and may contribute up to ~80% of the functional hyperemia response (6). Thus, the emerging view suggests that pericytes also contribute to blood flow regulation under physiologic and pathologic conditions, which should be investigated in greater detail by future studies.

### Phagocytosis and clearance of extracellular molecules

Pericytes take up multiple soluble small molecules, for example, horseradish peroxidase, india ink and dextran, via non-specific pinocytosis irrespective of route of administration—including peripheral injection, intraventricular injection and direct introduction in brain extracellular fluid [reviewed in (41, 162)]. Engulfed molecules are transported to lysosomes for enzymatic degradation (41, 162). In chronic BBB disruption models, pericyte phagocytic properties help clear toxic circulating plasma proteins that are normally excluded from the brain, including immunoglobulins, fibrin and albumin (5, 14). In models of acute brain injury, pericytes phagocytose cellular debris (24, 103). Recently, neuroinflammation has been shown to accelerate pericyte phagocytic activity (130). In addition to non-specific uptake, pericytes may also help specifically regulate the neuronal microenvironment by handling clearance of certain macromolecules in both physiologic and pathologic conditions. For example, pericytes express both the Aβ clearance receptor low-density lipoprotein receptor-related protein-1 (LRP1) and the ATP-binding cassette protein ABCA1 and may therefore contribute to brain Aβ and cholesterol homeostasis, respectively (139, 142, 175). Thus, pericyte degeneration may result in accumulation of different metabolites in the CNS that are normally cleared by pericytes.

## VASCULAR MECHANISMS IN AD

AD is the most common cause of dementia worldwide and is characterized by three histopathologic hallmarks—accumulation of brain and vascular Aβ (133), tau hyperphosphorylation and formation of neurofibrillary tangles (10, 77), and neuronal loss (133, 194). Epidemiologic studies have identified considerable overlap between risk factors for cerebrovascular disease and AD (76, 81, 165). The presence of cerebral hypoperfusion (137), subclinical infarcts (170) and/or the presence of one or more cerebral infarcts increase risk for AD (152). Conversely, control of vascular risk factors reduces vascular lesions in AD and may delay disease progression (40, 93, 135). Most AD pathologic specimens have evidence of mixed vascular pathology and small vessel disease with vascular changes being found in >40% of AD pathologic specimens (80, 98, 145, 163). Pathologic changes include pericyte loss and degeneration (11, 139, 146), atrophy and degeneration of vSMC, endothelial loss and reduction in capillary density, formation of collapsed and acellular capillary tubes (so-called string vessels), mitochondrial alterations, a thickened vascular basement membrane, loss of tight junctional and adherens junction proteins, increased endothelial pinocytosis and vesicular transport and BBB compromise leading to accumulation of plasma proteins in brain parenchyma, ISF and cerebrospinal fluid (CSF) [reviewed in (21, 50, 75, 196)].

Although classically defined by the amyloid hypothesis (65), these findings have raised questions as to the role of Aβ in AD and have led to the development of a vascular two-hit hypothesis of AD pathogenesis (Figure 2) (140, 141, 194, 196). This hypothesis maintains that reduced CBF gives rise to hypoxia and chronic perfusion stress, from one end, and BBB disruption gives rise to brain accumulation of plasma-derived neurotoxins, from the other, which may converge and initiate neuronal dysfunction and degeneration independently and/or prior to Aβ deposition and the development of tau pathology (81, 141, 196). Vascular dysfunction and injury also impairs clearance of Aβ from the brain (39, 199), leads to increased influx of circulating Aβ into the brain (37, 47) and elevates expression and/or processing of the Aβ precursor protein (APP) (2, 189). This results in Aβ accumulation and deposition within brain parenchyma and surrounding cerebral blood vessels. Aβ may then accelerate both vascular (12, 139, 196) and neuronal injury (173, 186) and promote self-propagation of cerebral and vascular β-amyloidosis (47, 105). The following subsections describe some mechanisms through which microvascular pathology contributes to AD-like neurodegeneration. The integral role of the pericyte in mediating vascular and neuronal changes in AD is described at length in a separate subsection (Figure 3) (please see the section Pericytes in AD).

### BBB disruption in AD

In human subjects, post-mortem brain tissue studies have shown that disruption of the BBB is associated with AD neuropathology and cognitive impairment (31, 33, 34, 51, 53, 57, 63, 74, 124, 138, 143, 146, 195, 196). The time point at which AD disruption occurs during disease pathogenesis remains unclear. Recent studies in human subjects at genetic risk for AD, such as carriers of the apolipoprotein ε4 allele (ApoE4), suggest that vascular permeability changes occur prior to cognitive decline (61). In transgenic mice overexpressing human APP, BBB permeability changes precede neuronal injury and AD pathology (139, 168). The mechanisms of BBB disruption remain poorly defined but may result from vasculotoxic effects of pericyte loss and/or endothelial injury

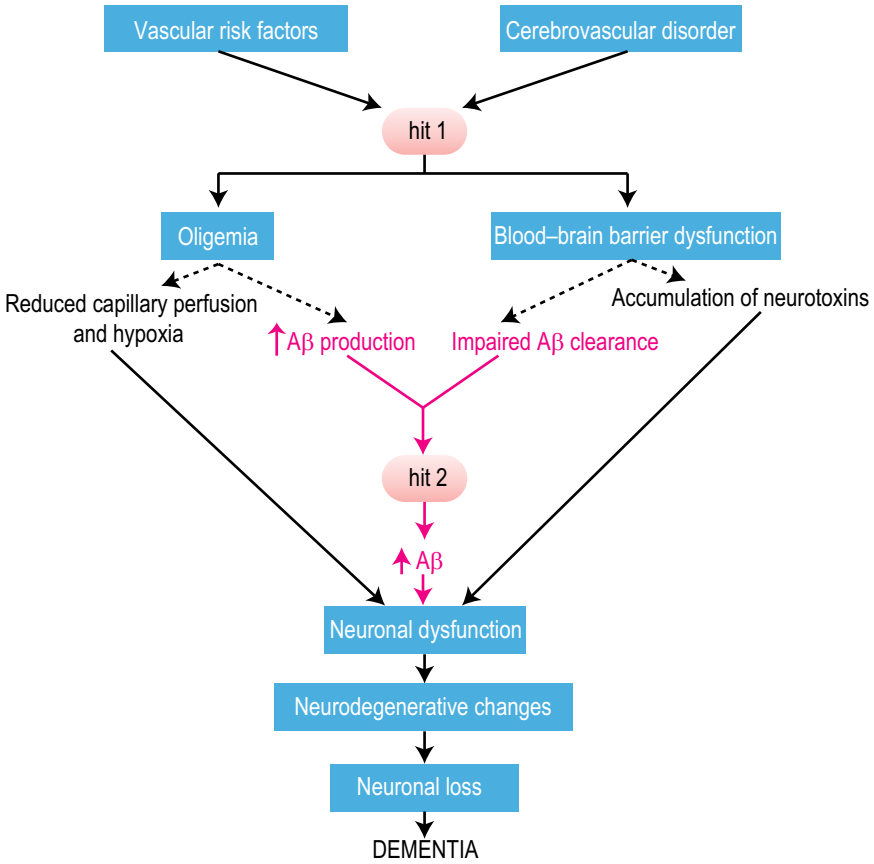


**Figure 2.** The vascular two hit hypothesis of Alzheimer's neurodegeneration. Vascular injury as result of long-standing vascular risk factors, for example, hypertension, dyslipidemia, diabetes, smoking, or obesity, genetic risk, for example, apolipoprotein ε4, and/or other unidentified environmental or toxic injury leads to early vascular dysfunction characterized by abnormalities in endothelial cells, pericytes and vascular smooth muscle cells. Vascular cell dysfunction and/or degeneration results in hypoperfusion (oligemia) and blood–brain barrier (BBB) dysfunction (*hit 1*) leading to hypoxia and accumulation of multiple plasma-derived neurotoxins, respectively, and contributes to neuronal dysfunction, degeneration and development of cognitive decline (solid lines). BBB dysfunction, mural cell loss and hypoperfusion/hypoxia reduce vascular amyloid β-peptide (Aβ) clearance across the BBB and in vascular mural cells and increases production of Aβ from Aβ-precursor protein (APP), causing Aβ accumulation in brain (*hit 2*, dashed lines). Pathologic elevations in soluble Aβ lead to the formation of neurotoxic Aβ oligomers and accelerates Aβ deposition around neurons and on blood vessels. Aβ species then amplify both vascular and neuronal injury. Tau phosphorylation and/or pathology, for example, caspase-cleavage and/or formation of neurofibrillary tangles, within neurons results from convergence of vascular injury (hypoperfusion and BBB disruption) and direct Aβ neurotoxicity. Figure modified from Zlokovic (196).

as a result of vascular risk factors, that is, dyslipidemia and/or Aβ (19, 139, 146, 168). Heightened vascular permeability may arise from reduced expression or accelerated degradation of tight and adherens junctional proteins, increased bulk-flow transcytosis, overt vascular rupture and/or combination thereof (195, 196). In transgenic mice expressing human ApoE4, increased activation of matrix metalloproteinase-9 degrades tight and adherens junctional proteins leading to vascular leakage (15). A similar cascade has been identified in human ApoE4 carriers (61). Whether a similar pathogenic cascade is erroneously activated in sporadic AD in the absence of ApoE4 remains to be experimentally determined.

Irrespective of the mechanisms involved, the end by-product of vascular disruption is accumulation of circulating plasma proteins and erythrocyte-derived hemoglobin in brain ISF and parenchyma (177, 196). Extravasated proteins may remain in the extracellular compartment or become internalized by different cell types of the NVU, including microglia, pericytes and neurons, and frequently co-localize with markers of cell death and/or injury (5, 14, 15, 101, 178). In multiple animal models, BBB disruption is an important contributor to neuronal injury and dysfunction (14, 15, 139, 178, 190). The mechanisms of injury are numerous. For example, brain accumulation of thrombin has been demonstrated in AD (192), and elevated thrombin concentrations lead to neuronal and vascular injury as well as cognitive impairment (26, 106). Plasmin degrades neuronal laminin, thereby disrupting neuron-extracellular matrix and sensitizing neurons to secondary injury (27). Fibrin and/or its precursor fibrinogen propagate vascular injury and neuroinflammation through accelerated vascular regression and barrier disruption (124). Aβ interacts with and stabilizes fibrin clots (32), and greater fibrin deposition is seen in vessels inflicted with cerebral amyloid angiopathy (CAA) (74). Depletion of fibrinogen reduces CAA and cognitive decline in transgenic AD mice (32) and may

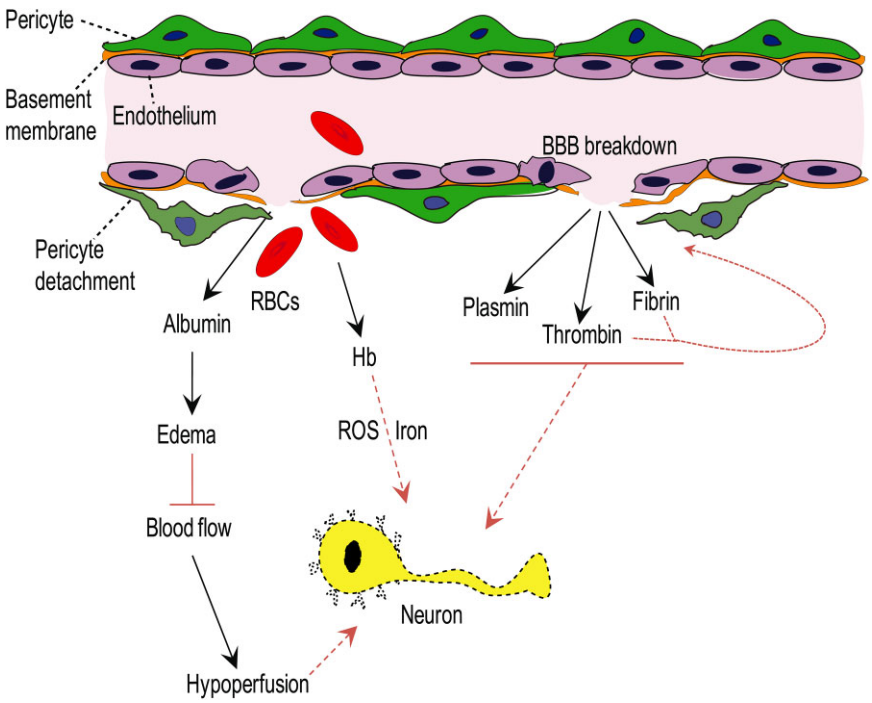


**Figure 3.** Pericyte degeneration leads to a chronic BBB disruption and vascular-mediated secondary neuronal injury and degeneration. Pericyte loss and/or degeneration represent an important cellular source of hit 1 vascular injury in Alzheimer's disease. Pericyte degeneration leads to BBB disruption and unrestricted entry and accumulation of blood-derived products in brain including erythrocyte-derived hemoglobin and plasma-derived proteins such as albumin, plasmin, thrombin, fibrin, immunoglobulin and others. Plasmin and thrombin have direct neurotoxic properties, whereas fibrin accelerates neurovascular injury. Brain degradation of hemoglobin liberates free iron, which catalyzes formation of reactive oxygen species (ROS) leading to further injury. Albumin increases oncotic pressure resulting in edema, microvascular compression and reduced blood flow. Pericyte loss also leads to endothelial cell death and microvascular regression leading to additional simultaneous reductions in blood flow. In mouse models, vascular injury in absence of Aβ as a result of pericyte loss is sufficient for neurodegeneration. Figure modified from Zlokovic (196).

therefore represent an important therapeutic target. In addition to plasma proteins, extravasation of erythrocytes due to microhemorrhage causes deposition of hemoglobin-derived products, such as free iron, which generates reactive oxygen species and non-specific oxidant injury (182, 190, 191). The presence of these proteins as well as albumin and immunoglobulin G contributes to brain edema and may alter perfusion dynamics through compression of brain capillaries (195, 196). Finally, disruption of the BBB may increase brain influx of circulating Aβ (37, 146).

## Endothelial dysfunction

Abnormal endothelial function leads to excess generation of potentially neurotoxic and pro-inflammatory molecules (58). For example, AD microvessels secrete neurotoxic factors, such as thrombin, which injure and kill neurons (59, 188). Isolated microvessels from human AD subjects also secrete and/or express a number of pro-inflammatory mediators, such as nitric oxide, cytokines (tumor necrosis factor, interleukin-1β and interleukin-6), chemokines (monocyte chemoattractant protein 1 and interleukin-8), prostaglandins and leukocyte adhesion molecules (58). In addition to neurotoxic and/or pro-inflammatory properties, AD endothelial cells demonstrate abnormal metabolic functions (99, 100). The BBB prevents unregulated influx of circulating polar solutes, including glucose, from blood into brain. Therefore, expression of endothelial GLUT1, which is also known as the solute carrier family 2, facilitated glucose transporter membrane 1 (SLC2A1), is required for facilitated diffusion of glucose into the brain. In human AD, endothelial GLUT1 protein levels are reduced (70, 82, 107). Abnormalities in glucose transport and/or utilization are visualized early with 2-[$^{18}$F] fluoro-2-deoxy-D-glucose positron emission tomography imaging (FDG-PET) in the AD hippocampus, parietotemporal cortex and/or posterior cingulate cortex (127). In those at genetic risk for AD, abnormal glucose handling precedes brain atrophy and neuronal dysfunction (108, 132). However, the significance of these findings remains controversial, and whether reduced glucose transport contributes to or, conversely, is the by-product of ongoing neurodegeneration and resulting reductions in metabolic demand still awaits the final answer.

## Altered vascular Aβ clearance

Pathologic accumulation of Aβ oligomers and insoluble fibrils may theoretically result from overproduction and/or diminished brain clearance of β-amyloid. In sporadic AD, however, impaired Aβ clearance, but not overproduction, is the key factor leading to Aβ

accumulation (102). Aβ may be cleared through transendothelial trafficking to circulating blood, enzymatic degradation such as neprilysin, insulin-degrading enzyme, tissue plasminogen activator and MMPs, in astrocytes, vSMC, pericytes and neurons within parenchyma, and clearance through perivascular ISF-CSF bulk flow (16, 196, 199). The vascular pathway is the major route of removal of Aβ from the brain (25, 149, 199). The predominate clearance protein—low-density lipoprotein receptor-related protein-1 (LRP-1)—binds Aβ on its abluminal plasma membrane and initiates its clearance to blood, as shown in multiple animal models (13, 39, 78, 149) and *in vitro* BBB models (39, 110, 184). ApoE4, but not ApoE2 or ApoE3, significantly inhibits LRP1-mediated Aβ brain clearance (38). Apolipoprotein J, also known as clusterin, contributes to Aβ clearance via a related receptor—the low-density lipoprotein receptor-related protein-2 (LRP-2) (13). The efflux transporter P-glycoprotein also makes contributions to vascular β-amyloid clearance (29). In addition to transendothelial transport, LRP1 expression in both vSMC and pericytes mediate Aβ uptake and degradation in vascular mural cells (12, 84, 139, 175). Reductions in LRP1 levels and/or alterations in receptor function as a result of oxidation are associated with brain Aβ accumulation in rodents, non-human primates, transgenic AD mice and human subjects with AD (9, 12, 39, 43, 149).

### Brain hypoperfusion in AD

Neurons are exquisitely sensitive to reductions in CBF—called "hypoperfusion". In human subjects and animal models, hypoperfusion is sufficient to induce cognitive decline (17, 76, 196). At a cellular level, moderate (~20%) blood flow reductions may result in altered protein synthesis impairing processes, such as long-term potentiation and synaptic plastic, required for learning and memory (71, 75). At greater reductions (>30%), the synthesis of ATP becomes impaired and anaerobic metabolism prevails. This results in reduced ATPase activity limiting generation of action potentials and cellular transport, thus contributing to neuronal dysfunction and/or alterations in the neuronal microenvironment, that is, lower cellular pH, electrolyte imbalances and cytotoxic edema (140, 196). Acute reductions in CBF (>80%) result in ischemic neuronal death (109).

Reductions in CBF have not only been observed in human AD subjects (8, 75, 104, 196), but in some studies precede neurodegenerative changes (75, 85, 86, 137, 147, 151). In APOE4 carriers, CBF reductions and reduced CBF responses to brain activation may occur prior to brain atrophy and Aβ accumulation and worsen as the disease progresses (85, 147). A similar relationship between blood flow changes and course of disease has been demonstrated in transgenic mouse models expressing both human APOE4 and APP (15, 112, 139). A number of studies in animal models suggest that reduced CBF accelerates multiple stages of AD neurodegeneration. In transgenic AD mouse models, bilateral occlusion of the common carotid artery increases neuronal Aβ levels and tau phosphorylation and accelerates cognitive decline and neuronal loss (87, 185). In human AD and transgenic models, hypoperfusion appears to accelerate vascular amyloid deposition and CAA, which may, in turn, lead to further ischemia and microinfarctions (116). Therefore, hypoperfusion and subsequent Aβ accumulation may constitute a feed-forward loop.

AD-related impairment in blood flow may result from at least two processes: aberrant vascular reactivity and microvascular degeneration. In human subjects, CBF changes occur prior to changes in vascular volume as evidenced by cerebral blood volume magnetic resonance imaging (MRI) (91). This suggests that abnormal vascular function may precede vascular degeneration. The mechanism(s) through which vascular reactivity becomes disrupted in AD have been shown to involve pathologic contractile properties in vSMC in small penetrating arteries. For example, studies in both human AD subjects and transgenic models have demonstrated upregulation of two transcription factors, myocardin (MYOCD) and serum response factor (SRF), important for controlling vSMC differentiation and leads to upregulation of vSMC contractile proteins, a hypercontractile vSMC phenotype, and, ultimately, impaired blood flow (12, 28). In human AD, reduced microvascular diameter suggestive of a hypercontractile phenotype has been observed in the hippocampus (22). MYOCD and SRF overexpression in microvascular pericytes has also been suggested (R.D. Bell, E.A. Winkler and B.V. Zlokovic, unpub. data) and may contribute to these changes. However, further investigation is needed to define the role of contractile protein expression in AD pericytes.

Aβ may also lead to vessel contraction and impairment in CBF as evidenced by the vaconstrictory effect of exogenous Aβ (111, 113, 114, 119) and aberrant vascular reactivity in transgenic models overexpressing APP (112, 139, 164, 183). Aβ impairs CBF on both luminal and abluminal targets (123). On the luminal membrane, Aβ binding to the receptor for advanced glycation end products (RAGE) leads to production of endothelin, a potent a potent vasoconstrictor (37, 195). Meanwhile, Aβ binding to the scavenging receptor CD36 on vascular and perivascular cells leads to oxidative stress and associated vascular dysfunction (121, 122). Deletion of CD36 protects against CBF reductions, vascular Aβ accumulation and slows cognitive decline in aged mice overexpressing APP (122). Therefore, improvement in CBF may delay AD histopathologic and behavior changes.

In addition to dysfunctional vasomotor responses, histologic studies have demonstrated focal vascular regression and reduced focal vascular density in human AD and transgenic models (11, 21, 58, 120, 139, 183). Neuroimaging studies demonstrating reductions in cerebral blood volume corroborate findings of microvascular degeneration (91, 167, 181). Genome-wide transcriptional profiling in AD human endothelium has identified downregulation in the mesenchyme homeobox gene-2 (MEOX2), a transcription factor important for vascular differentiation and patterning, as a key contributor to aberrant angiogenic responses to angiogenic stimuli, endothelial apoptosis and chronic hypoperfusion (183). In mouse models, heterozygosity for *Meox2* is sufficient to induce neurodegenerative changes (14). Contributions of additional sources of vascular injury have been documented, including pericyte loss (11, 139, 146), Aβ vasculotoxicity (161) and/or Aβ-mediated inhibition of angiogenesis (120). Whether pericyte loss precedes and/or leads to downregulation of endothelial *Meox2* remains to be experimentally determined.

## PERICYTES IN AD

Multiple independent reports have demonstrated pericyte loss and/or degeneration in both the hippocampus and cortex in human AD subjects (11, 50, 146). At an ultrastructural level, pericytes

demonstrate large numbers of intracellular inclusions, pinocytotic vesicles, large lipid granules and mitochondrial abnormalities, suggesting cellular dysfunction and/or degeneration (11, 50). Pericyte degenerative changes are associated with capillary reductions and gross dilatation and tortuosity of surviving vessels (11). In individual AD subjects, reductions in pericyte coverage inversely correlate with evidence of BBB disruption, such as leakage of the plasma proteins including immunoglobulin G and fibrin (146). Although these observations are limited to post-mortem tissue, pericyte dysfunction and/or loss are associated with key attributes of AD vascular pathology—vascular regression and disrupted vascular permeability.

## Factors contributing to pericyte loss in AD

The mechanism(s) of pericyte loss have yet to be completely defined. Some studies have suggested that brain pericytes do not decrease with normal aging in rodents during adulthood (14, 125, 128). Additional studies in normally aged animals are needed, however, to confirm these findings. Preliminary data have suggested that vascular factors, for example, hypertension and dyslipidemia, may lead to pericyte injury and/or death (153, 158). Future works are still needed to establish a contributory link between vascular injury and early pericyte loss in AD. In later disease stages, Aβ accumulates on and around brain capillaries and pericytes (171, 180). Pericytes express the Aβ clearance receptor LRP-1 which binds and internalizes different Aβ species for lysosomal degradation within brain pericytes (139, 175). At high concentrations and prolonged exposure, Aβ species overwhelm the clearance capacity and lead to pericyte cell death *in vitro* (139, 175). Reductions in pericyte cell number and/or coverage have been similarly observed in transgenic mice with β-amyloidosis *in vivo* as a result of overexpression of human APP and accumulation of Aβ in pericytes (122, 139). Other works have also implicated the scavenging receptor CD36 in AD-related pericyte loss (122).

A loss of brain pericytes, in turn, leads to reduced Aβ clearance through the LRP-1 degradative pathway promoting Aβ accumulation and/or deposition in the brain (139). Therefore, pericytes are important for brain Aβ clearance but are susceptible to Aβ toxicity, a potential feed-forward mechanism. In human AD subjects, extravascular Aβ deposits inversely correlate with pericyte coverage in the AD hippocampus—meaning those with greatest Aβ load have fewest pericytes (146). This finding does not permit directionality of this relationship to be established but may reflect the by-product of both Aβ-driven pericyte toxicity and a loss of pericyte-dependent Aβ clearance.

## Pericytes and Aβ-independent neurodegeneration

Recent works have suggested that a loss and/or dysfunction of brain pericytes may accelerate AD-like neurodegeneration cascade *in vivo*. In mouse models, a loss of brain pericytes is sufficient to induce neurodegenerative changes in the absence of Aβ through two major pathways—BBB disruption and hypoperfusion (Figure 3) (14, 177, 178). On the one hand, a loss of pericytes leads to heightened vascular permeability through a disrupted BBB. This, in turn, leads to an accumulation of multiple blood-derived neurotoxic and vasculotoxic molecules seen in human AD, including fibrin (124), thrombin (26, 106), plasmin (27) and hemoglobin-derived iron and reactive oxygen species (73, 134, 182). Concomitantly, a loss of pericytes also leads to regression of brain microvessels, mainly capillaries. This gives rise to a chronic perfusion stress and hypoxia. Toxin accumulation and hypoxia may then simultaneously converge at the neuronal interface resulting in injury, dysfunction and ultimately cell death (14).

The importance of both pathways to the neurodegenerative phenotype was demonstrated in comparative studies between two different mouse lines: a mouse deficient in PDGFRβ signaling (*Pdgfrβ*$^{F7/F7}$ mice) and a mouse heterozygous for mesenchyme homeobox gene-2 involved in vascular patterning (*Meox2*$^{+/-}$mice). As previously mentioned, *Meox2* is transcriptionally downregulated in AD and leads to reduced angiogenesis, microvascular reductions and diminished CBF (183). *Meox2*$^{+/-}$ mice have reduced microvascular density, but intact pericyte populations and normal BBB integrity (14, 183). In contrast, *Pdgfrβ*$^{F7/F7}$ mice have a deficiency in brain pericytes, which leads to both a microvascular reduction and a disruption. In both mouse lines, neurodegeneration was observed, suggesting that chronic hypoperfusion is sufficient to induce neuronal injury. However, the magnitude of such changes was much larger in pericyte deficient mutants highlighting the synergy between blood-derived toxin accumulation and hypoxia.

## Pericytes and APOE genotype

Further evidence for the importance of Aβ-independent vascular injury in neurodegenerative changes comes from works in two transgenic mouse lines overexpressing the human *APOE4* gene (15, 197). The three major apoE isoforms (E2, E3 and E4) differ in single amino acid substitutions at two distinct residues and alter the structure and function of apoE at molecular and cellular levels (197). *APOE4* is the strongest genetic risk factor for sporadic AD (160). Individuals homozygous for *APOE4* have a 30% and 60% lifetime risk to develop disease at 75 and 85 years of age, respectively (55). The *APOE4* gene may contribute to significant neurovascular dysfunction prior to cognitive decline and Aβ-accumulation (194, 197) and is associated with greater deficits in BBB permeability in AD (74, 143, 192).

The mechanism of these BBB permeability changes was not established until recently. In mice, overexpression of human apoE4, but not apoE2 or apoE3, leads to upregulation of a pro-inflammatory cyclophilin A (CypA)-nuclear factor κB (NF-κB)-matrix metalloproteinase 9 (MMP-9) pathway within brain pericytes (Figure 4). MMP-9 is then secreted from pericytes and becomes activated. This, in turn, leads to enzymatic degradation of endothelial cell tight junctional complexes resulting in BBB breakdown and unregulated influx of the blood-derived neurotoxins, including thrombin, fibrin and hemoglobin-derived iron. Chronic barrier disruption in addition to caspase-independent effects of apoE4 led to pericyte reductions, microvascular regression and CBF reductions. Importantly, apoE4-driven vascular injury was sufficient to induce neuronal dysfunction, injury and ultimately cell death. Pharmacologic interventions inhibiting each step of the CypA-NFκB-MMP-9 pathway with specific inhibitors of CypA and NF-κB or inhibition of MMP-9, and genetic deletion of CypA reduced both vascular and neuronal dysfunction in transgenic *APOE4* mice, suggesting that dysfunction of pericytes can be

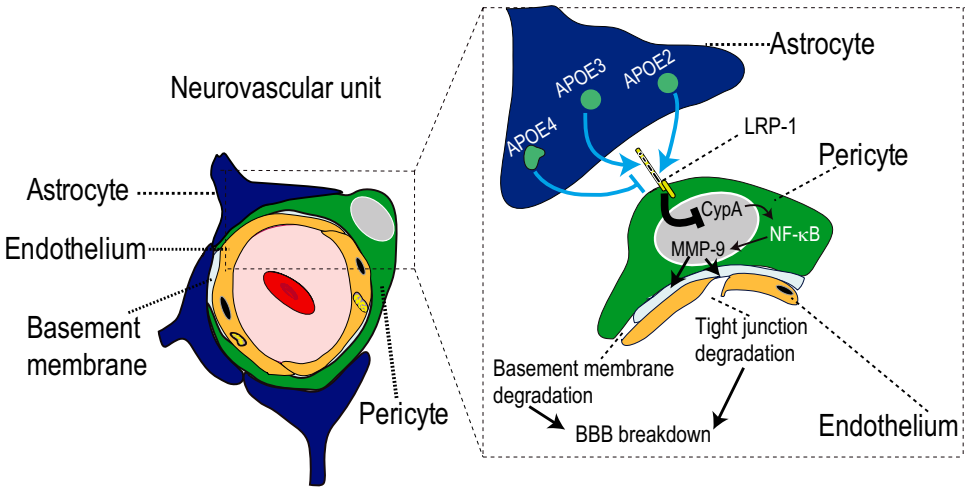


**Figure 4.** Apolipoprotein E4 triggers early pericyte dysfunction and blood–brain barrier (BBB) breakdown. Apolipoproteins are secreted by astrocytes. ApoE2 and apoE3, but not apoE4, bind to the low density lipoprotein receptor-related protein-1 (LRP-1) on pericytes and suppress the levels of a pro-inflammatory cytokine cyclophilin A (CypA). ApoE4 exhibits weak binding to LRP-1 resulting in pathologic elevations in CypA protein levels, which, in turn, increases nuclear translocation of the pro-inflammatory transcription factor nuclear factor-κB (NF-κB), and upregulation of pericyte matrix metalloproteinase-9 (MMP-9). Secretion and activation of MMP-9 degrades endothelial tight and adherens junction proteins leading to disruption of the BBB. Pathologic elevations of MMP-9 and a disrupted BBB lead to capillary loss and hypoperfusion. Vascular injury may then lead to neuronal dysfunction and degeneration. Figure modified from Bell *et al* (15).

therapeutically controlled and that pericytes may represent a novel cellular therapeutic target (15).

This work has recently been extended to cognitively normal subjects including *APOE4* carriers and *APOE4* non-carriers (61). This study has shown that individuals harboring the *APOE4* allele develop an age-dependent BBB breakdown as evidenced by elevated CSF : plasma albumin quotient, which correlated with increased CypA levels and activated MMP-9 levels in the CSF, suggesting that activation of the pro-inflammatory CypA pathway occur before cognitive decline (61). Additionally, a recent study reported that CypA mRNA levels are reduced in *APOE2* carriers further supporting the role of CypA in regulating neurovascular function (30). Collectively, these data suggest that the BBB damage likely involving pericyte dysfunction might contribute to early neurovascular dysfunction seen in animal models and humans with the *APOE4* allele.

### Pericytes and Aβ accumulation

To determine whether a loss of brain pericytes also contributes to Aβ-dependent toxicity in AD-like neurodegeneration, our group recently published a study in which pericyte deficient mutants (*Pdgfrβ*$^{+/-}$ mice) were crossed with mice which overexpressing the Swedish mutation of human APP (*APP*$^{sw/0}$) (139) (Figure 5). *APP*$^{sw/0}$ mice accumulate parenchymal and vascular Aβ leading to amyloid plaques and memory deficits, but do not develop tau pathology and/or neuronal loss (72, 154). Enhanced pericyte loss (*APP*$^{sw/0}$*;Pdgfrβ*$^{+/-}$ mice) leads to greater accumulation of soluble $A\beta_{40}$ and $A\beta_{42}$ species as a result of reduced pericyte-dependent clearance of soluble Aβ species from brain ISF, as shown by *in vivo* microdialysis in the hippocampus. Impaired clearance of soluble Aβ then contributes to accelerated deposition of insoluble Aβ resulting in CAA and parenchymal β-amyloid plaques.

### Pericytes and tau pathology

*APP*$^{sw/0}$*;Pdgfrβ*$^{+/-}$ mice also developed tau pathology at the early age of 9 months including neuronal accumulation of hyperphosphoryled tau species, caspase-cleaved tau and tau aggregates (139). Importantly, tau pathology at this relatively early disease stage was not observed in either *APP*$^{sw/0}$ mice or *Pdgfrβ*$^{+/-}$ mice, suggesting that both pericyte-driven vascular injury and Aβ elevations must be present to induce early appearance of tau pathology. With respect to neuronal loss, *Pdgfrβ*$^{+/-}$ mice did show a more moderate neuronal loss as a result of direct vascular injury as previously described (14). However, the magnitude of neuronal dysfunction and/or loss, and behavioral impairment on hippocampal-dependent tasks was much more severe in *APP*$^{sw/0}$*;Pdgfrβ*$^{+/-}$ mice (139). Consistent with the two-hit vascular hypothesis, pericyte-driven direct vascular injury and Aβ working together can induce tau pathology and neuronal loss *in vivo* amplifying cognitive decline. Aβ accumulation in pericytes in turn leads to pericyte loss, worsening AD-related neurovascular and neuronal dysfunction (Figure 5).

## SUMMARY AND FUTURE PERSPECTIVES

The contributions of pericytes to neurodegenerative disease have only recently been recognized. Among chronic neurodegenerative diseases, pericyte loss and/or dysfunction have been implicated in AD pathogenesis. However, pericyte degeneration seems not to be unique to AD as pericyte loss and dysfunction have been recently reported in other neurodegenerative disorders including ALS (179) and cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy (CADASIL) (60), as well as in

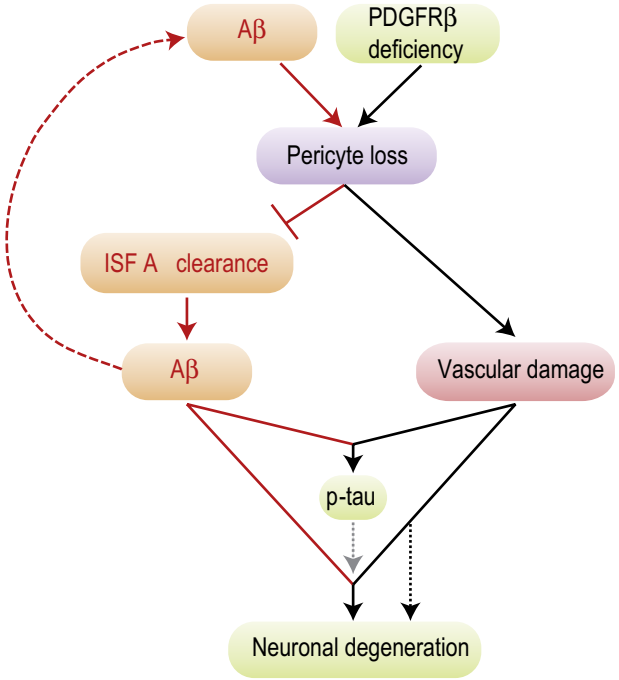


**Figure 5.** Pericyte degeneration and loss contributes to Alzheimer's type neurodegeneration through amyloid β-peptide (Aβ) independent and dependent injury mechanisms. Brain pericyte loss as a result of disruption of the platelet derived growth factor receptor β (PDGFRβ) signaling triggers early Aβ-independent vascular injury leading to microvascular loss and disruption, which results in hypoperfusion/hypoxia and brain accumulation of toxic plasma proteins, respectively (black). Loss of pericytes also leads to diminished clearance of soluble Aβ species from brain interstitial fluid (ISF) and further elevations in brain Aβ levels (red). Aβ may then overwhelm degradative pathway in surviving pericytes, resulting in further pericyte degeneration (dashed line). Both pericyte loss and Aβ acting simultaneously result in the early development of all facets of Alzheimer's disease neuropathology including Aβ plaques and neuronal tau pathology, degeneration and loss, which are not observed at an early disease stage when Aβ accumulation or pericyte-driven vascular injury occurs in isolation. Figure modified from Sagare *et al* (139).

transgenic models and individuals carrying APOE4, a major genetic risk factor for AD (15, 61).

As shown in transgenic models of pericyte deficiency and AD, pericytes may contribute to disease pathogenesis through both Aβ-independent pathway and by altering Aβ metabolism and clearance. Both pathways may act synergistically after disease onset to amplify microvascular degeneration, causing neuronal dysfunction and degeneration. Whether pericyte injury and degeneration represents a common downstream pathway for neurodegenerative changes in AD has yet to be explored by using models with vascular risk factors alone (eg, hypertension, diabetes, dyslipidemia, homocysteinemia and pro-coagulant profile) and/or in combination with models of Aβ and tau pathology. It would also be interesting to investigate whether pericytes play a role in the metabolism and clearance of other proteinaceous material implicated in the pathogenesis of other neurodegenerative diseases including Parkinson's disease (eg, α-synuclein), prion disease and/or ALS (eg, superoxide dismutase).

Preliminary studies have supported that pericytes may be therapeutically targeted to stabilize peripheral vascular lesions (94). Therapies focused on CNS pericytes have yet to be developed experimentally and tested for chronic neurodegenerative disease and/or brain vascular lesions. Initial attempts have been made by showing successfully that either genetic or pharmacologic inhibition of the pro-inflammatory CypA pathway in brain pericytes in rescues both the vascular and the neuronal phenotype in APOE4 transgenic mice (15). Future studies are needed, however, to extend these findings to humans with *APOE4* genetic risk. Similarly, more work is needed to identify other key molecular players and pathways mediating aberrant signal transduction between pericytes, endothelial cells, astrocytes and neurons within the NVU in sporadic and inherited AD that could possibly represent new therapeutic targets to control pericyte degeneration in dementia and slow vascular and neuronal degeneration. Only then may pericyte-targeted therapies find broader applications for neurological disorders and lead to the development of new neurovascular medicine.

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